

International Application No.: PCT/AU03/00696

International Filing Date: June 4, 2003

U.S. Application No.: 10/517,003

Response to Notification of Missing Requirements and Preliminary Amendment

Amendments to the Claims:

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims:

1. (Original) A solid phase comprising a surface first chemical moiety which participates in covalent bond formation with a second chemical moiety conjugated to a tag oligonucleotide rendered partially double stranded by annealing an α-tag oligonucleotide to the tag oligonucleotide to provide a 3' overhang portion of the tag oligonucleotide wherein the tag oligonucleotide is employed as a substrate for ligase-mediated covalent bonding to a target nucleic acid molecule.

2. (Original) The solid phase of Claim 1 comprising a solid support in the form of a microsphere, microchip or a glass, plastic or ceramic slide.

3. (Original) The solid phase of Claim 2 wherein the solid support is a microsphere.

4. (Original) The solid phase of Claim 1 wherein the surface chemical moiety is capable of covalent bond formation with an amine group, a thiol group or an acryl group.

5. (Original) The solid phase of Claim 1 wherein the surface chemical moiety is capable of covalent bond formation with a carboxyl group.

6. (Original) The solid phase of Claim 1 wherein the surface chemical moiety is a carboxyl group.

7. (Original) The solid phase of Claim 1 wherein the second chemical moiety is an amine group.

8. (Original) The solid phase of Claim 1 wherein the tag oligonucleotide comprises a chemical moiety conjugated to a known oligonucleotide sequence *via* a molecule comprising $mc+n$ atoms, from about 1 to about 100, wherein m is the number of repeats of size C and n is the number of atoms not included in the repeats.

9. (Original) The solid phase of Claim 1 wherein the α -tag oligonucleotide is labeled with a reporter molecule and is phosphorylated at its 5' end.

10. (Currently Amended) The solid phase of Claim 8 or 9 further comprising a bridging oligonucleotide, said bridging oligonucleotide having a nucleotide sequence complementary to the nucleotide sequence of the 3' overhang portion of the tag oligonucleotide and a nucleotide sequence complementary to a terminal end portion of a target nucleic acid molecule.

11. (Original) The solid phase of Claim 10 further comprising a target nucleic acid molecule in ligase-mediated covalent bonding to the tag oligonucleotide molecule anchored to the solid phase.

12. (Original) A substrate for anchoring a target nucleic acid molecule, said substrate comprising:

- (i) a solid phase having a first chemical moiety on its surface;
- (ii) a tag oligonucleotide comprising a second chemical moiety in covalent bond formation with the first chemical moiety, said second chemical moiety conjugated to the tag oligonucleotide *via* a molecule of structure $mc+n$ from about 1 to

International Application No.: PCT/AU03/00696

International Filing Date: June 4, 2003

U.S. Application No.: 10/517,003

Response to Notification of Missing Requirements and Preliminary Amendment

about 100, wherein m is the number of repeats of size c and n is the number of atoms not included in the repeats.;

(iii) an optionally labeled α -tag oligonucleotide complementary to the tag oligonucleotide resulting in a 3' singled-stranded overhang of the tag oligonucleotide; and

(iv) a bridge oligonucleotide having complementary based to the 3' overhang region of the tag oligonucleotide and complementary bases to the 5' end portion of the target nucleic acid molecule wherein the target nucleic acid molecule is anchored to the tag oligonucleotide *via* ligase-mediated conjugation.

13. (Canceled)

14. (Original) A universal nucleic acid anchoring system comprising the structure:



wherein:

S is a solid support having a chemical moiety capable of covalent bond formation with a second chemical moiety;

T is a partially double-stranded oligonucleotide comprising a single-stranded tag oligonucleotide having said second chemical moiety linked *via* a spacer molecule to its 5' end, said spacer comprising carbon atoms having the structure mc+n from about 1 to about 100, wherein m is the number of repeats of size c and n is the number of atoms not included in the repeats, said tag oligonucleotide further comprising a complementary oligonucleotide (α -tag) annealed to the tag oligonucleotide to provide a 3' overhang or sticky end, single-stranded nucleotide sequence, on the tag oligonucleotide; said T further comprising a bridging oligonucleotide having a nucleotide sequence complementary to the 3' overhang nucleotide sequence on the tag

International Application No.: PCT/AU03/00696

International Filing Date: June 4, 2003

U.S. Application No.: 10/517,003

Response to Notification of Missing Requirements and Preliminary Amendment

oligonucleotide and a further nucleotide sequence complementary to a nucleotide sequence on the 5' end of a target nucleic acid molecule;

wherein T may be represented p times on the solid support wherein p is from about 1 to about 100,000.

15. (Original) A method for immobilizing a target nucleic acid molecule to a partially double-stranded tag oligonucleotide anchored to a solid support, said method comprising ligating a phosphorylated 5' end of the target nucleic acid molecule to a complementary single-stranded portion of the tag oligonucleotide under conditions to permit ligase-mediated covalent bond formation wherein said tag oligonucleotide is covalently anchored to the solid support *via* covalent bond formation between a first chemical moiety on the surface of the solid support and a chemical moiety conjugated to the tag oligonucleotide *via* a molecule of structure $mc+n$ from about 1 to about 100, wherein m is the number of repeats of size c and n is the number of atoms not included in the repeats wherein the tag oligonucleotide is rendered partially double-stranded by annealing a complementary oligonucleotide to the tag oligonucleotide leaving a single-stranded 3' terminal portion of the tag oligonucleotide which is used to capture the target nucleic acid molecule *via* a bridging oligonucleotide.

16. (New) The solid phase of Claim 9 further comprising a bridging oligonucleotide, said bridging oligonucleotide having a nucleotide sequence complementary to the nucleotide sequence of the 3' overhang portion of the tag oligonucleotide and a nucleotide sequence complementary to a terminal end portion of a target nucleic acid molecule.